

Camel (*Camelus dromedarius*) Mastitis in Borena Lowland Pastoral Area, Southwestern Ethiopia

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Key words

Camelus dromedarius – Mastitis –
Somatic cell count – Lactation –
Ethiopia.

Summary

Quarter-milk samples (n = 828) from 207 traditionally managed lactating camels (*Camelus dromedarius*) in Borena (Southwestern Ethiopia) were examined to determine the occurrence and bacterial causes of mastitis in the camel. The California mastitis test (CMT) was used as a screening test and bacteriological examinations were carried out to identify the mastitis pathogens involved. Somatic cell counts (SCC) of camel milk samples were also determined. Out of 828 camel quarters examined 25 (12.1%) teats were blind. An agreement of 100% was found for CMT scores of 3+ and 2+ and bacteriological results, while 35, 71 and 85% agreements were observed for CMT scores of 0, trace and 1+, respectively. A significant association was observed between CMT positive scores and the presence of major pathogens in camel milk samples. SCC ranged from 3×10^5 to 1.5×10^7 leukocytes/ml of milk. An increasing number in the mean values of somatic cell counts was obtained for increasing scores of CMT using ANOVA. Four (1.9%) of the lactating camels examined were detected as clinical cases of mastitis. Among the CMT positive quarter-milk samples examined, 171 (74%) yielded pathogenic bacteria. The major mastitis pathogens isolated included species of *Staphylococcus*, *Streptococcus*, *Micrococcus*, *Corynebacterium* and *Bacillus*, and *Actinomyces pyogenes*, *Escherichia coli* and *Pasteurella haemolytica*.

INTRODUCTION

The dromedary camel is a multipurpose animal adapted to the harsh environments of semiarid and arid zones, essentially kept for milk and meat production and transportation. It is also a financial reserve (asset) and security (drought-prone risk management) for pastoralists and plays an important role in social prestige and wealth (8, 19). Because of the increasing desertification and recurrence of drought and famine in sub-

Saharan Africa, particularly in East Africa, the camel plays a very significant role as a source of milk, meat and draft power. In Ethiopia camels are kept in the arid and semiarid lowlands of Borena, Ogaden and Afar regions, which cover 50% of the pastoralist areas in the country. The major ethnic groups owning camels in Ethiopia are the Somali, Borena and Afar (21).

Milk of camel is one of the main components of the diet of the nomads in Ethiopia and is consumed in its raw or naturally processed (soured) form (21). Very little work has been done on mastitis in the camel as the disease was thought to be uncommon in this species (1). However, during the past years mastitis in the camel has been reported from a number of camel-rearing countries of the world (1, 2, 3, 7, 10, 17). The present study was undertaken to find out the occurrence and bacterial causes of camel mastitis in the camel in Borena, Southwestern Ethiopia (Figure 1). The performance of the California mastitis test (CMT) as a screening test for the detection of mastitis in the camel was evaluated. The relationship between udder infection and somatic cell counts (SCC) was also determined.

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Figure 1: Map of Ethiopia with Borena province.

■ MATERIALS AND METHODS

A total of 207 lactating she-camels from four villages in Liben district (Borena), Southwestern Ethiopia, raised by nomadic tribes were examined. There were 104 she-camels from Kersemele and Koradisa villages and 103 from Boba and Bulbuli villages in the first and second visits, respectively.

Clinical examination of udder and milk

Udder abnormalities such as swelling, presence of lesions or anatomical malformations were recorded. The size of the rear and forequarters, indurations and fibrosis were examined by deep digital palpation. Tick infestations and use of antisuckling devices were also noted. The milk was examined for its consistency, color and other visible abnormalities. Clinical mastitis was recognized by abnormal milk, signs of udder infection and detection of mastitis pathogens by bacteriological culture, whereas subclinical mastitis was recognized by apparently normal milk and an increase in leukocyte counts as evidenced by CMT and a positive culture result.

Milk sample collection

Before milk sampling, the teats were disinfected with cotton moistened with 70% alcohol. After discarding the first few squirts of milk about 20 ml were collected in sterile universal bottles and kept in an icebox, and transported immediately to the laboratory for analysis. Out of 828 quarters examined, 25 (12.1%) teats were blind. Therefore, a total of 803 quarter-milk samples were collected and used for analysis.

California mastitis test

CMT was used according to the procedure described by Schalm *et al.* (18). CMT score 0 was taken as negative, while CMT scores trace, 1+, 2+ and 3+ were considered positive, thus forming five categorical classes. All milk samples irrespective of CMT results were bacteriologically examined.

Somatic cell counts

SCC were carried out to establish the relationship between the udder infection and the number of cells in camel milk. The direct microscopic somatic cell count (DMSCC) method as described by Packard *et al.* was used (12).

Isolation and identification of bacteria

Bacteriological examinations were carried out following standard methods (9, 14, 20). Briefly, a loopful of each milk sample was streaked on 7% sheep blood agar (Merck). MacConkey agar (Merck) plates were also used in parallel to detect *Enterococcus* species and any Gram-negative bacteria. Inoculated plates were incubated aerobically at 37°C for 24-48 h. Presumptive identification of bacterial isolates was made based on colony morphologic features, Gram-stain reaction, hemolytic characteristics and a catalase test. *Staphylococci* and *Micrococci* were identified based on their growth characteristics on mannitol salt agar, coagulase production, catalase and oxidase tests. Isolates identified tentatively as *Streptococci* were evaluated according to CAMP reaction, growth characteristics on Edward's medium (Oxoid), hydrolysis of esculin and sodium hippurate, catalase production, and sugar fermentation tests. Gram-negative isolates were subcultured on MacConkey agar and further tested using triple sugar iron (TSI) agar (Merck), the IMViC test (indole, methyl red, Voges-Proskauer and citrate utilization tests), urea, lysine and ornithine decarboxylase and oxidase reactions.

■ RESULTS

The majority of camel udders examined were infested with ticks (*Amblyomma*, *Hyalomma* and *Rhipicephalus* species). The teat/udder skin lesions were superficial and old. Twenty-five (12.1%) of the 207 dromedary udders examined had atrophied and blind teats. From a total of 207 lactating camels examined, 4 (1.9%) were clinical cases of mastitis. Clinically affected udders were swollen, hard and painful upon palpation. Camel owners in Borena use antisuckling devices to prevent calves from suckling. For this purpose, they use bark of a tree as a string to tie up pairs of teats together. This is done during daytime when young camels older than one year are herded together with their dams. Table I shows CMT and SCC values and the types of bacteria isolated from clinical cases of mastitis.

Out of 803 quarter-milk samples tested with CMT, 231 (28.8%) were positive. Out of these 231 CMT positive samples, 171 (74%) yielded pathogenic bacteria on culturing. Some of these samples contained bacteria, which gave mixed isolations. A significant agreement percentage was observed between CMT scores and bacteriological findings (Table II). Of the CMT positive milk samples, 26% (60/231) yielded no bacterial growth.

CMT scores of 2+ (n = 9) and 3+ (n = 2) samples showed a 100% agreement percentage for bacteriological results while CMT scores

Table I

California mastitis test (CMT), somatic cell counts (SCC) and bacteria isolated from clinical cases of mastitis in camels

CMT score	SCC (cells/ml)	Bacteria isolated
1+	3×10^5	<i>Staphylococcus aureus</i> <i>Staph. hyicus</i>
2+	1.9×10^6 6.8×10^6	<i>Streptococcus agalactiae</i> <i>Staph. aureus</i>
3+	4.4×10^6	<i>Strept. agalactiae</i>

Table II

Relation between the California mastitis test (CMT) scores and bacteriological results of camel milk samples

CMT score	Num. of samples tested	Culture		% agreement
		Positive	Negative	
0	572	200	372	35
T	187	132	55	71
1+	33	28	5	85
2+	9	9	0	100
3+	2	2	0	100
Total	803	371	432	

CMT score (positive = trace, 1+, 2+, 3+, versus negative 0) and culture results (positive versus negative) with significant association, Chi square $P < 0.001$

of trace (T) and 1+ showed 71 and 85% agreements, respectively. In order to evaluate CMT scores with respect to the importance of the organism isolated associated with mastitis, the bacteria were classified as major pathogens (MAP) if *Streptococcus agalactiae*, *Staphylococcus aureus*, *Escherichia coli*, *Strep. uberis*, *Strep. dysgalactiae*, *Enterococcus faecalis*, *Actinomyces pyogenes*, *Corynebacterium ulcerans* and *Pasteurella haemolytica* were isolated, and they were classified as minor pathogens (MIP) if *Corynebacterium bovis*, *Staph. epidermidis*, *Staph. hyicus*, *Staph. intermedius* and *Bacillus* species were isolated (14, 15, 16).

A statistically significant association ($P < 0.001$) was observed between CMT (when categorizing CMT positive T and all + against negative 0) and cultures (MAP, MIP aggregated into positive cultures) using a chi square test (Table II).

The somatic cell counts of 803 milk samples ranged from 3×10^5 to 1.5×10^7 leukocytes/ml milk. The overall leukocyte count for NI (non-infected), MIP and MAP bacterial groups indicated that there was a decreasing trend in the range of SCC, respectively. Because of the skewed distribution of SCC among quarter-milk samples examined, SCC was log transformed. It was observed that quarters infected with MAP had higher mean values of SCC than MIP groups and non-infected quarters (Table III). The somatic cell counts for MAP were significantly higher ($p < 0.01$) than those for MIP and NI. Table IV shows the means of SCC (log scale) with respect to categories of CMT scores. Trend of means given categories of CMT is presented in Figure 2.

Gram-positive cocci were dominant among the total of 818 bacteria isolated from camel milk samples (Table V).

DISCUSSION

The udder/teat skin lesions observed in lactating camels could be attributed to the tick burden (*Amblyomma*, *Hyalomma* and *Rhipicephalus* species) infesting the udders and scratches caused by thorny plants of the desert. The use of antisuckling devices in Borena camels is practiced only during daytime when young calves older than one year are herded together with their dams. The use of these devices together with heavy tick infestation could predispose the udders to bacterial infections, which persist as chronic infections. This could result in induration and atrophy of injured quarters (11). Other non-traumatic devices such as the ones

Table IV

Mean of somatic cell counts (SCC) (log scale) for categories of California mastitis test (CMT) scores

CMT	Mean Log SCC	Standard Deviation	Num. of observations
0	2.78	2.57	577
Trace	4.02	2.31	182
1+	3.47	2.74	33
2+	5.27	2.04	9
3+	6.05	0.86	2

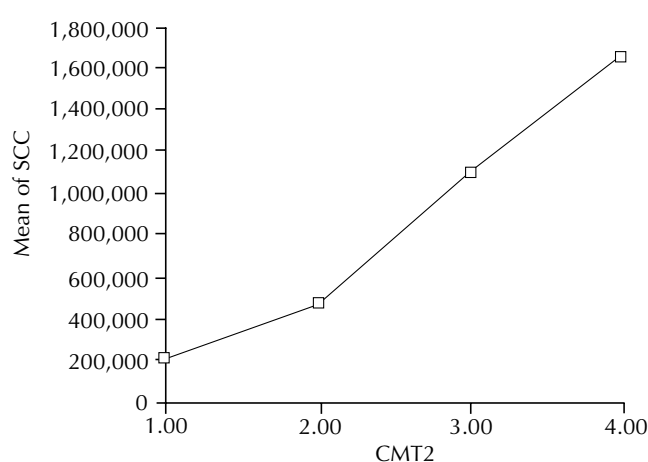


Figure 2: Trend of SCC means given four categories of CMT ($p < 0.001$, ANOVA SPSS, with CMT scores 2+ and 3+ considered together).

Table III

Mean of somatic cell counts (SCC) for major pathogens (MAP), minor pathogens (MIP) and non-infected (NI) quarter groups

	Num. of observations	MAP (n = 202)	MIP (n = 324)	NI (n = 89)
Log SCC	615	12.45 \pm 0.105	11.87 \pm 0.073	11.51 \pm 0.116

Table V

Mastitis pathogens isolated from camels with clinical and subclinical mastitis

Mastitis pathogens	Num. of cases		Total number (%)
	Clinical mastitis	Subclinical mastitis	
<i>Staphylococcus aureus</i>	2	171	173 (21.1)
<i>Staph. hyicus</i>	1	206	207 (25.3)
<i>Streptococcus dysgalactiae</i>	-	28	28 (3.4)
<i>Strept. agalactiae</i>	2	24	26 (3.2)
<i>Enterococcus faecalis</i>	-	18	18 (2.2)
<i>Strept. uberis</i>	-	7	7 (0.9)
<i>Micrococcus</i> spp.	-	86	86 (10.5)
<i>Staph. intermedius</i>	-	67	67 (8.2)
<i>Staph. epidermidis</i>	-	81	81 (9.9)
<i>Corynebacterium ulcerans</i>	-	15	15 (1.8)
<i>Corynebacterium bovis</i>	-	13	13 (1.6)
<i>Actinomyces pyogenes</i>	-	5	5 (0.6)
<i>Escherichia coli</i>	-	3	3 (0.4)
<i>Pasteurella haemolytica</i>	-	1	1 (0.1)
<i>Bacillus</i> spp.	-	88	88 (10.8)
Total	5	813	818

used in Mauritania, a protecting harness made of rope named "Shmell" (4), could serve alternatively to reduce injury incidence. In addition to camels, nomads in Borena keep other animals such as cattle and goats, which could be sources of mastitis pathogens. Other species of animals, which are kept together with camels, could serve as sources of udder infection (11).

The strong positive correlation of CMT with the bacteriological findings indicates that camel milk, like that of cows (18), goats and sheep (5), has phagocytic cells that constitute one of the essential defenses against microbial infections. Milk samples infected with MAP had higher CMT scores. CMT can be used as a screening test to detect subclinically infected udders of camels (3).

The results of the somatic cell counts of the milk samples (between 3×10^5 and 1.5×10^7 cells/ml) is in agreement with that of Kospakove (1976), cited by Abdurahman *et al.* (1), who reported a mean of 1.3×10^6 cells/milk sample in Bactrian camels. The range of SCC was very small for the MAP group as compared to the MIP and NI groups. Udders infected with MAP had higher values with CMT, SCC and adenosine triphosphate (ATP) than samples from MIP and NI quarters with somatic cell counts of 7.4×10^5 – 1.2×10^7 cells/ml (1). An increase in the number of somatic cells in camel milk with infected quarters has also been reported by Mostafa *et al.* (10). The possibility that NI quarters have a high SCC can also be supported by the fact that most of the examined camels had lesions on the teats/udder skin because of the string used to tie the teats to prevent suckling or because of ticks.

The rate of milk samples reacting positively with CMT but yielding no bacterial growth was lower (28.2%) than that of Abdurahman *et al.* (1). They reported that 43% of CMT-positive quarter-milk samples of camels did not show any bacterial growth

on appropriate media. This may be because the samples were taken during the convalescent phase of the infection but with a high leukocyte count giving a CMT-positive result. *Escherichia coli* and other coliforms tend to be of very short duration as they are rapidly destroyed by inflammatory reactions. It has also been reported that 57% of coliform infections usually last less than 10 days and milk samples can be culture negative in 20% of clinical cases of mastitis (14, 20). Milk samples from cows with clinical mastitis or from cows whose milk has high somatic cell counts often yield no organism on culture (5). In other cases, the infection may have been eliminated, but an elevated SCC persists still infiltrating the udder tissues.

An increasing trend of mean of SCC and log scale of mean of SCC for CMT scores was observed even though this lacks equal distribution of the frequency of the SCC between the different CMT scores. An increase in milk cell counts also reflects positive CMT results. In the present study, both SCC and CMT results showed they were valuable indicators of udder infections in the camel. An increase in the mean values of both CMT and SCC was observed for the MAP bacterial group as compared to the MIP and NI groups.

The instantaneous prevalence of mastitis in the sample of the study (4/207) was relatively lower than that observed by other investigators elsewhere (1, 2, 3, 11, 13). Gram-positive cocci were the main pathogens isolated from camel milk samples constituting 84.7% of the total isolates. Various authors also reported that these pathogens are major mastitis causing agents in camels (1, 3, 6, 7, 10, 11), in dairy cows (5, 14, 15, 16, 18), and in goats and sheep (5, 6). Agents found in positive cultures, like *Staphylococcus aureus* and other species of *Staphylococcus*, were mainly

responsible for subclinical mastitis, but some agents, like *Streptococcus agalactiae*, were found in both clinical and subclinical mastitis as already described by Younan *et al.* (22). They reported that *Strept. agalactiae* and *Staph. aureus* were found in 12.1 and 10.6% of camel milk samples examined in Kenya, respectively, whereas in the present study 3 and 21% were found, respectively. The increased number of isolates of *Strep. agalactiae* and other major mastitis pathogens could be attributed to the lack of supply and infrequent use of antimicrobials, and the inaccessibility of the camel owners to veterinary services as compared to dairy cow owners in urban and periurban areas.

■ CONCLUSION

Results of the present study showed that mastitis was prevalent in dromedary camels of the Borena zone of Southwestern Ethiopia,

and Gram-positive cocci were the dominant mastitis pathogens isolated. The positive correlation of CMT with the presence of mastitis pathogens in camel milk showed that CMT is a useful screening test in the detection of mastitis in camels and may serve to segregate udders infected with major pathogens in a subclinical form. Increase in the somatic cell counts of infected quarters indicated that camels reacted to inflammation induced by agents of mammary tissue by raising the number of somatic cells in the milk. However, further investigation is needed to determine the infection threshold of SCC in camel milk.

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Résumé

Woubit S., Bayleyegn M., Bonnet P., Jean-Baptiste S. Mammmites du dromadaire (*Camelus dromedarius*) dans la région pastorale basse du Borana au sud-ouest de l'Éthiopie

Du lait de quartier (n = 828) a été prélevé chez 207 femelles dromadaires (*Camelus dromedarius*) en lactation, provenant de troupeaux du Borana, au sud-ouest de l'Éthiopie, et élevées de manière traditionnelle. L'objectif de l'étude a été de décrire la prévalence des mammites et certaines étiologies bactériennes chez le dromadaire. Le California mastitis test (Cmt) a été utilisé comme test de dépistage et des examens bactériologiques ont été effectués pour identifier les agents pathogènes impliqués dans les mammites. La numération cellulaire somatique du lait de quartier des chamelles a également été déterminée. Vingt-cinq quartiers (12,1 p. 100) ont été trouvés non productifs parmi les 828 examinés. Un pourcentage de correspondance de 100 p. 100 a été trouvé pour les échantillons classés 3+ et 2+ avec Cmt, alors qu'un pourcentage de correspondance de 35, 71 et 85 p. 100 a été relevé pour ceux classés respectivement 0, traces et 1+ avec Cmt. Une association significative a été observée dans le lait de quartier des chamelles entre les classements positifs obtenus avec Cmt et la présence d'agents pathogènes principaux. La numération cellulaire somatique a été comprise entre 3×10^5 et $1,5 \times 10^7$ leucocytes par millilitre de lait. Les moyennes du comptage cellulaire ont montré une évolution numérique positive en fonction des classes croissantes du Cmt avec Anova. Parmi les femelles en lactation examinées, quatre (1,9 p. 100) cas cliniques de mammites ont été détectés. Des bactéries pathogènes ont été présentes dans 171 échantillons (74 p. 100) de lait de quartier examiné positif avec Cmt. Parmi les principaux agents pathogènes isolés ont été trouvées des espèces de *Staphylococcus*, *Streptococcus*, *Micrococcus*, *Corynebacterium* et *Bacillus*, ainsi que *Actinomyces pyogenes*, *Escherichia coli* et *Pasteurella haemolytica*.

Mots-clés : *Camelus dromedarius* – Mammite – Numération cellulaire somatique – Lactation – Éthiopie.

Resumen

Woubit S., Bayleyegn M., Bonnet P., Jean-Baptiste S. Mastitis en camellas (*Camelus dromedarius*) en la zona pastoril baja de Borena, sudoeste de Etiopía

Se examinaron muestras de leche (n = 828) de 207 camellas (*Camelus dromedarius*) lactantes, bajo manejo tradicional, en Borena (sudoeste de Etiopía), con el fin de determinar la presencia y las causas bacterianas de mastitis en la camella. Se utilizó el California Mastitis Test (CMT), como test de despiste y se llevaron a cabo exámenes bacteriológicos para la identificación de los patógenos involucrados con la mastitis. Se obtuvieron también conteos de células somáticas (SCC) en las muestras de leche de camella. De los 828 cuartos de camella examinados, 25 (12,1%) pezones fueron ciegos. Una concordancia de 100% fue encontrada para los valores de CMT de 3+ y 2+, con los resultados bacteriológicos; mientras que una concordancia de 35, 71 y 85% fue observada con los valores del CMT de 0, traza y 1+, respectivamente. Se observó una asociación significativa entre los valores positivos del CMT y la presencia de los patógenos importantes en las muestras de leche de camella. Los SCC variaron de 3×10^5 a $1,5 \times 10^7$ leucocitos/ml de leche. Se obtuvo un número creciente en los valores promedio de los conteos de células somáticas, con respecto a valores crecientes del CMT mediante el uso de ANOVA. En cuatro (1,9%) de las camellas lactantes examinadas fueron detectados casos clínicos de mastitis. Entre los CMT positivos de los cuartos de leche examinados, 171 (74%) mostraron bacterias patógenas. Los principales patógenos de mastitis aislados incluyeron especies de *Staphylococcus*, *Streptococcus*, *Micrococcus*, *Corynebacterium* y *Bacillus*, y *Actinomyces pyogenes*, *Escherichia coli* y *Pasteurella haemolytica*.

Palabras clave: *Camelus dromedarius* – Mastitis – Conteo de células somáticas – Lactación – Etiopía.